

Takaaki KOBARA* & Mitsuo CHIHARA*: **On the life history
of *Pseudobryopsis hainanensis* (Chlorophyceae)**

高原隆明*・千原光雄*: 緑藻ハネモドキの生活史について

Introduction

It was shown in our previous paper (Kobara & Chihara, 1978) that *Pseudobryopsis hainanensis* Tseng¹⁾ collected from Amami Islands produced male and female gametes and the zygotes developed on germination into prostrate non-septate filaments with constrictions at places and with branches sparsely and irregularly. For obtaining further knowledges concerning the life history of this alga, we have continued to culture in the laboratory the filaments derived from the germinating zygotes. As a result, the life history has been completed *in vitro* starting from both zygotes and zoospores. The present paper gives an account of asexual reproduction and life history of *Pseudobryopsis hainanensis* Tseng cultured under laboratory conditions.

Materials and Methods

The specimens used in the present study were collected from northern region of Amami Islands, at Ankyaba on March 25 and 26, 1978, and at Sani on March 27, 1978. Morphological features of the prostrate filaments derived from the germinating zygotes in culture were given in detail in our previous paper (Kobara & Chihara, 1978). Outline of the culture methods is also fundamentally the same as that described in the previous paper. However, for inducing the formation of zoospores in the filaments, the following two sets of culture conditions were employed in addition to the previously used condition (set 1)²⁾: set 2—temperature, 27°C; photoperiod (hr light : hr dark), 14:10, and set 3—temperature, 23°C; photoperiod, 10:14. These conditions were selected to correspond approximately to intertidal conditions in the southern regions of Japan, including Amami Islands. The set 1 and set 2 were maintained at a light intensity of 4000–6000 lux from 30W cool white

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¹⁾ *Trichosolen hainanensis* (Tseng) Taylor, according to Taylor (1962).

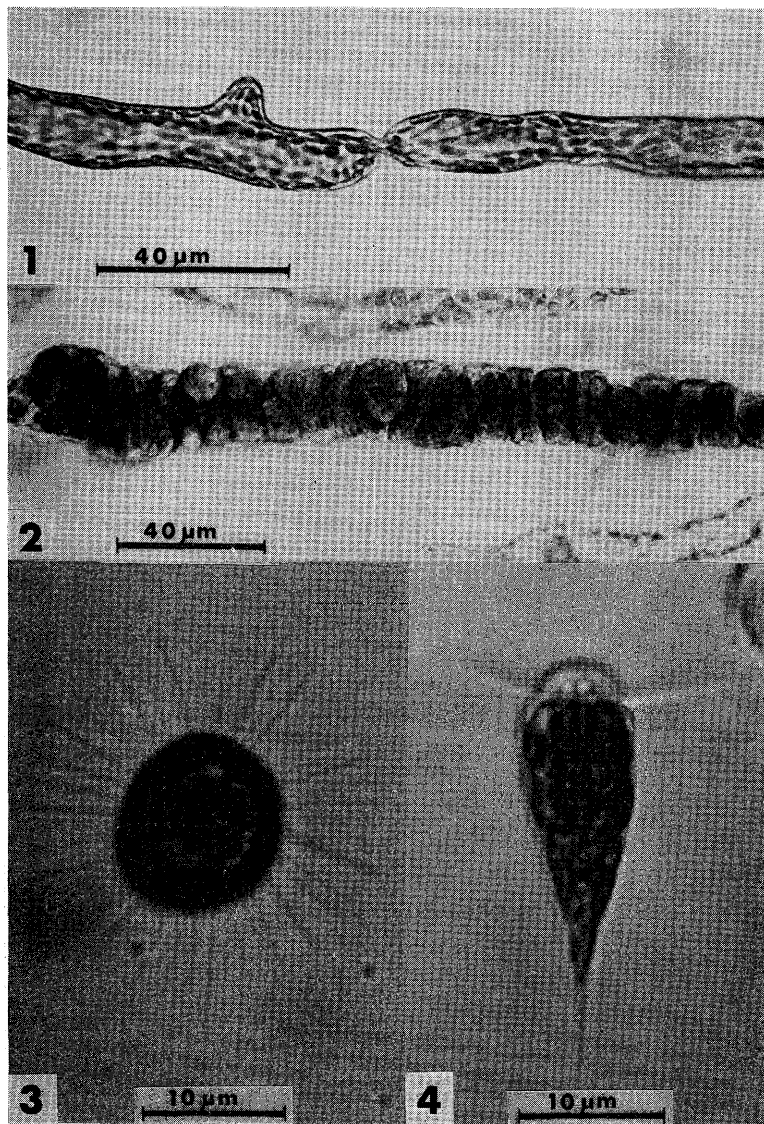
²⁾ Temperature, 20°C; photoperiod (hr light : hr dark), 14:10.

fluorescent lamps, while the set 3 was at 3000-4000 lux.

Observations

The filamentous germlings cultured under the set 1 condition continued to grow keeping their filamentous states (Fig. 1) and never developed into an erect axis similar to that found in nature. In order to induce the formation of asexual cells, the filaments cultured in this condition were transferred to the set 2 condition and maintained there for about three weeks. Then, they were transferred to the set 3 condition, whose culture was aerated by air pump. After about two days in this culture, the formation of asexual cells took place in the filaments. Manner in which the zoospores were formed was as follows. The whole content of the filament becomes dense in color, and the chloroplasts, nuclei and some other cellular structures become aggregated, resulting in the formation of a large pack of protoplasm which is somewhat homogeneous and densely dark. It occupies almost entire parts inside the filament, except for the distal parts. Soon, the content gives rise to numerous zoospores, undergoing divisions which take place simultaneously with one another and almost perpendicularly or somewhat obliquely to the longer axis of the filament (Fig. 2). These zoospores are usually produced in a transverse row in a slender filament, as is shown in Fig. 2, while in a thick filament, they lie partially one upon another though they form a row as a whole. Formation of zoospores thus occurred is in the manner of holocarpic and no specialized reproductive organs are produced. The zoospores are liberated one by one through an releasing aperture formed in wall at the terminal end of the filament. The zoospores are extremely characteristic in appearance, showing obovate to pyriform in shape, provided with a long posterior projection and many flagella which lie in a transverse whorl at the base of slightly protruding anterior end of the body (Figs. 3-5). They possess many chloroplasts, which occupy almost entire parts of the body, except for the very anterior and the tailed portions. The zoospores measure 280-330 μm long and 70-110 μm wide at the widest part of the body. They possess no eyespot.

After becoming attached to the substrates, zoospores lose flagella, become spherical (Fig. 6) and increase in volume, forming a vacuole at the central portion of the cell and increasing chloroplasts in number (Fig. 7). They begin to germinate pushing out from the upper portion of the cell a



Figs. 1-4. *Pseudobryopsis hainanensis*. 1. Portion of filamentous germling derived from the zygote. 2. Portion of the filament forming zoospores holocarpically. 3. A zoospore with many flagella lying in a transverse whorl at anterior end, viewed from anterior tip. 4. A zoospore with many flagella and a long posterior projection, viewed from lateral.

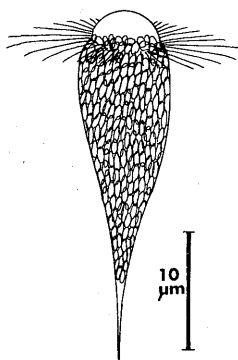


Fig. 5. Stephanokont zoospore of *Pseudobryopsis hainanensis*.

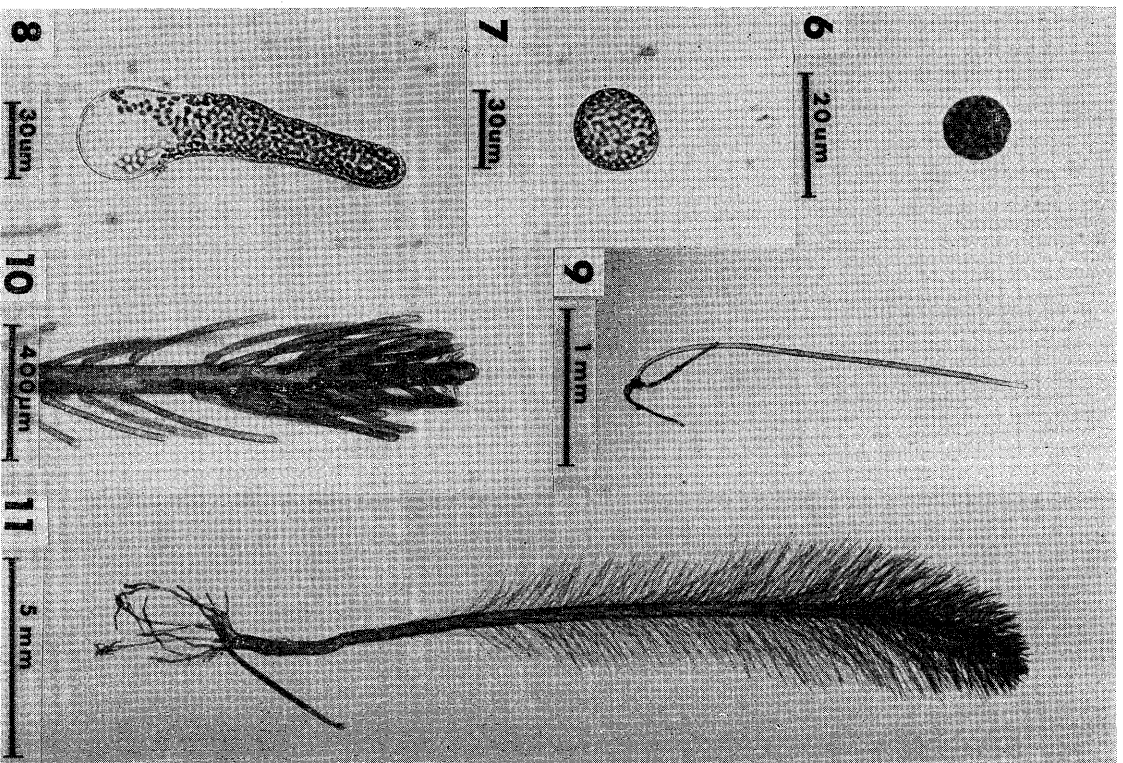
germ tube (Fig. 8), which later gives rise to an erect axis. The erect axis continues to grow towards the direction of light, producing rhizoidal filaments from the base, and becomes a long tubular upright filament (Fig. 9). The filament is coenocytic, and simple at beginning, but later produces many ramuli from all the sides in its upper part (Fig. 10). Several new axes are also produced from the rhizoidal filaments and they grow in the same manner as that of the initial upright filament, forming a tuft as a whole.

The germlings thus obtained in the laboratory culture were morphologically identical with the thalli of *Pseudobryopsis hainanensis* collected in nature (Fig. 11). After about four weeks in the culture of the set 1, the germlings reached 2.0-3.0 cm in height, became mature and formed many characteristic gametangia near the base of the ramuli. These gametangia produced gametes of both sexes, and the zygotes developed on germination into a prostrate filament similar to that described in our previous paper (Kobara & Chihara, 1978).

Discussion

It is apparent from the present investigation and results described in our previous paper that *Pseudobryopsis hainanensis* is a sexual phase, which produces male and female gametes, the zygotes develop into prostrate non-septate filaments, which produce very characteristic stephanokont zoospores, and the zoospores give rise on germination to the *Pseudobryopsis* phase (Fig. 12). These evidences indicate that the alga has an alternation of heteromorphic generations, in which a sexual *Pseudobryopsis* phase alternates with an asexual prostrate filamentous phase.

This type of life history agrees fundamentally with that found in certain members of *Bryopsis*, including the type species, *B. plumosa* (Rietema, 1969, 1975; Tatewaki, 1973). As far as is known, only one paper is available at present concerning the life history of *Pseudobryopsis*. It is of Mayhoub



Figs. 6-11. *Pseudoryopsis hainanensis*. 6. A zoospore attached on a slide glass (after 8 hours). 7. A settled zoospore increasing in volume and chloroplasts in number (after 24 hours). 8. Early stage of the germinating zoospore (after 3 days). 9. An upright germinating, having rhizoids at the base (after 8 days). 10. Portion of the upright germlings, having ramuli from all the sides of the axis. 11. *Pseudoryopsis* thallus derived from the germinating zoospore in culture.

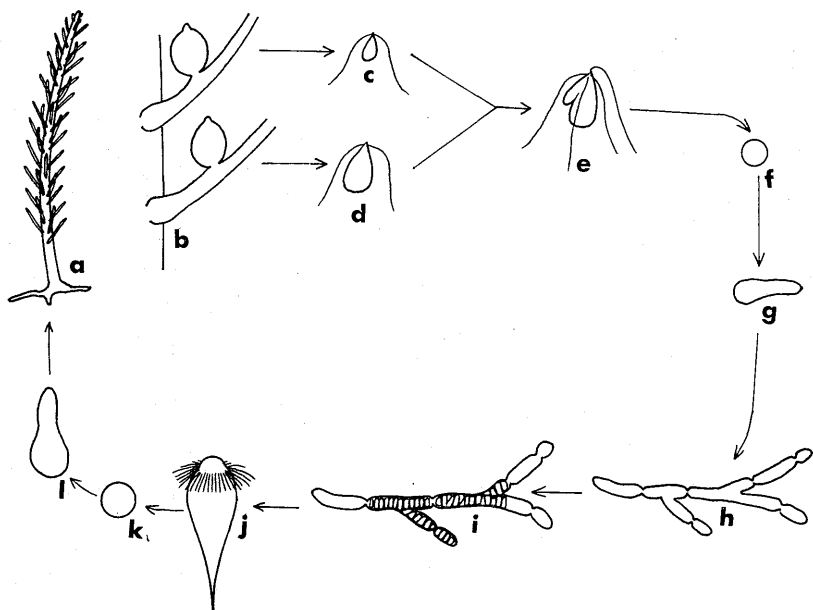


Fig. 12. Diagrammatic figure showing life history of *Pseudobryopsis hainanensis*. a, *Pseudobryopsis* thallus. b, male and female gametangia formed in the same thallus. c, male gamete. d, female gamete. e, conjugation of gametes. f, zygote. g, germination of zygote. h, filamentous germling derived from the zygote. i, filamentous germling forming zoospores holocarpically. j, stephanokont zoospore having a tail-like appendage. k, settled zoospore. l, germination of zoospore.

(1974) with *P. myura* (J. Agardh) Berthold³⁾ from the Mediterranean Sea. Our findings agree in some respects with Mayhoub's observations, but disagree in others. According to Mayhoub, the prostrate filaments from the germinating zygotes produce directly the *Pseudobryopsis* thalli and asexual reproduction by stephanokont zoospores is absent.

The genus *Bryopsis* and the related genera, including *Bryopsidella*, *Derbesia* and *Pedobesia*, have been the subjects of recent cultural studies by many workers, notably Rietema (1969, 1975) and Tatewaki (1973, 1977). As a result, it has been shown that there is a surprising diversity of life history types. On the basis of the information available, Rietema (1975) classified the life histories of this algal group into five types: 1) *Bryopsis-plumosa*

³⁾ *Trichosolen myura* (J. Agardh) Taylor, according to Taylor (1962).

(Zeeland) type, 2) *Bryopsis-plumosa* (Roscoff) type, 3) *Bryopsidella* type, 4) *Derbesia* type and 5) *Pedobesia* type. As already noted above, the life history of our alga belongs to the *Bryopsis-plumosa* (Roscoff) type, while that of Mayhoub's one belongs to *Bryopsis-plumosa* (Zeeland) type. It might be conceivable from these evidences that the genus *Pseudobryopsis* exhibits life histories that vary among the species as in the same way as *Bryopsis* and the related genera. With respect to the systematic position of *Pseudobryopsis*, our findings of the life history would lend support to the view that *Pseudobryopsis* is a genus related to *Bryopsis*. However, it might be worthwhile to examine biochemically the main constituents of cell walls of this algal group before accepting this general conclusion.

A striking feature of *Pseudobryopsis hainanensis* is the formation of obovate to pyriform stephanokont zoospores in the filaments derived from the germinating zygotes. Though the occurrence of stephanokont zoospores is well known in *Bryopsis* and the related genera, they are all spherical in shape, with somewhat protruding portion at anterior end of the body. In this connection, a comparative study of these zoospores is in progress with the aid of electron microscope.

Summary

The life history of *Pseudobryopsis hainanensis* collected from Amami Islands has been completed *in vitro* starting from both zygotes and zoospores. *Pseudobryopsis* thalli produce sexual cells, the zygotes develop into prostrate filaments, which later produce very characteristic stephanokont zoospores and the zoospores give rise on germination to *Pseudobryopsis* thalli similar to those found in nature.

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前報でわれわれ (高原・千原 1978) は奄美大島産のハネモドキの形態, 分類, 有性生殖, 接合子の発芽などについて報告した。本報告は前報に引続くもので, 無性生殖, 遊走子の発芽などについて培養により得た結果を記述した。20°C 長日条件で培養を続けた接合子からの発芽体を 27°C 長日条件に移して約 3 週間経た後に 23°C 短日条件で処理した処, 約 2 日後に遊走子の形成を見た。形成は全実的に起り, 遊走子は多鞭毛のいわゆる *stephanokont zoospore* で, 倒卵形に近く, 後端に尾状突起をもつ。この形状の遊走子は従来全く知られないタイプのものである。遊走子は直立型の発芽を行い, 天然に見られるハネモドキの体に発達する。生活史は有性世代のいわゆるハネモドキの体と無性世代の匍匐糸状の体の交代によることがわかる。

□日本ベゴニア協会編: 国内に導入されているベゴニア・リスト A check list of of *Begonia* species and hybrids introduced into Japan. pp. 58, 1978年8月. ベゴニア属は世界に約 2000種を産し, またそれらを親とした交配が多く行われて膨大な数に達している。それらの中から日本に到来したものを網羅したのが本目録である。これらを木立性, 根茎性, 球根, レックス, センパフフロレンス, 冬咲, エラチオール の 8 系統に分ち, 夫々を種や品種の ABC 順に配列し, その名の出典, 命名者, 発表年, 原産地, 性状, 品種では作者名, 発表年, ABC 登録番号を出来るだけ記載したものであって, その調査と取まとめに数年をかけている。その努力は大変なものであったと思うし, 担当された方々のお骨折りをしのぶ。一部600円, 送料200円で一般にも分かつという。

(前川文夫)